

Applicants: Ron S. Israeli et al.  
Serial No.: 08/466,381  
Filed: June 6, 1995

Pending Claims 97-103

97. A method of detecting micrometastatic prostate tumor cells in a subject which comprises:
- a) obtaining a suitable sample of mRNA from the subject;
  - b) contacting the mRNA sample under hybridizing conditions with a labeled nucleic acid probe which:
    - (1) is at least 15 nucleotides in length and (2) hybridizes specifically to a nucleic acid having a sequence which is complementary to a sequence present in the sequence set forth in SEQ ID NO. 1.
  - c) removing any unbound labeled nucleic acid probe; and
  - d) detecting the presence of labeled nucleic acid probe hybridized to the mRNA;
  - e) comparing the amount of labeled nucleic acid probe measured in step d) with an amount measured in a negative control sample which does not have micrometastatic prostate tumor cells, wherein a higher amount measured in step d) compared to the amount measured in the control sample indicates the detection of micrometastatic prostate tumor cells in the subject.
98. A method of detecting micrometastatic prostate tumor cells in a subject which comprises:
- a) obtaining a suitable sample of mRNA from the subject;
  - b) reverse transcribing the mRNA to generate a single-stranded cDNA;

Applicants: Ron S. Israeli et al.

Serial No.: 08/466,381

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- c) contacting the single-stranded cDNA under hybridizing conditions with a labeled nucleic acid probe which: 1) is at least 15 nucleotides in length; and 2) hybridizes specifically to a nucleic acid having a sequence set forth in SEQ ID NO:1;
  - d) removing any unbound labeled nucleic acid probe; and
  - e) detecting the presence of labeled nucleic acid probe hybridized to the cDNA;
  - f) comparing the amount of labeled nucleic acid probe measured in step e) with an amount measured in a negative control sample which does not have micrometastatic prostate tumor cells, wherein a higher amount measured in step e) compared to the amount measured in the control sample indicates the detection of micrometastatic prostate tumor cells in the subject.
99. A method of detecting micrometastatic prostate tumor cells in a subject which comprises:
- a) obtaining a suitable sample of mRNA from the subject;
  - b) generating a double-stranded mRNA-cDNA duplex from the mRNA;
  - c) contacting the duplex from (b) with one primer having a sequence which is complementary to a portion of the sequence set forth in SEQ ID NO:1 and a second primer having a sequence which comprises a different portion of the sequence set forth in SEQ ID NO:1;

Applicants: Ron S. Israeli et al.  
Serial No.: 08/466,381  
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- d) amplifying the nucleic acid from (c) using a polymerase chain reaction to obtain an amplification product;
- e) contacting the amplification product of (d) under hybridizing conditions with a labeled nucleic acid probe which: 1) is at least 15 nucleotides in length; 2) hybridizes specifically to a nucleic acid having a sequence set forth in SEQ ID NO. 1.;
- f) removing any unbound labeled nucleic acid probe; and
- g) detecting the presence of labeled nucleic acid probe hybridized to the amplification product;
- h) comparing the amount of labeled nucleic acid probe measured in step g) with an amount measured in a negative control sample which does not have micrometastatic prostate tumor cells, wherein a higher amount measured in step g) compared to the amount measured in the control sample indicates the detection of micrometastatic prostate tumor cells in the subject.

100. A method of detecting micrometastatic prostate tumor cells in a subject which comprises:

- a) obtaining a suitable sample of mRNA from the subject;
- b) generating a double-stranded mRNA-cDNA duplex from the mRNA;
- c) contacting the duplex from (b) with one primer having a sequence which is complementary to a portion of the sequence set forth in SEQ ID NO:1 and a second primer

Applicants: Ron S. Israeli et al.

Serial No.: 08/466,381

Filed: June 6, 1995

having a sequence which comprises a different portion of the sequence set forth in SEQ ID NO:1;

- d) amplifying the nucleic acid from (c) using a polymerase chain reaction to obtain an amplification product;
- e) contacting the amplification product of (d) under hybridizing conditions with a labeled nucleic acid probe which: 1) is at least 15 nucleotides in length; and 2) hybridizes specifically to a nucleic acid having a sequence complementary to the DNA sequence set forth in SEQ ID NO:1.;
- f) removing any unbound labeled nucleic acid probe; and
- g) detecting the presence of labeled nucleic acid probe hybridized to the amplification product;
- h) comparing the amount of labeled nucleic acid probe measured in step g) with an amount measured in a negative a control sample which does not have micrometastatic prostate tumor cells, wherein a higher amount measured in step g) compared to the amount measured in the control sample indicates the detection of micrometastatic prostate tumor cells in the subject.

101. A method of detecting the presence of a nucleic acid encoding a prostate specific membrane antigen in a subject which comprises:

- a) obtaining a suitable sample of mRNA from the subject;
- b) generating a double-stranded cDNA from the mRNA;

Applicants: Ron S. Israeli et al.  
Serial No.: 08/466,381  
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- c) contacting the double-stranded cDNA from (b) with one primer having a sequence which is complementary to a portion of the sequence set forth in SEQ ID NO:1 and a second primer having a sequence which comprises a different portion of the sequence set forth in SEQ ID NO:1;
- d) amplifying the double stranded cDNA using a polymerase chain reaction to obtain an amplification product;
- e) contacting the amplification product of (d) under hybridizing conditions with a labeled nucleic acid probe which 1) is at least 15 nucleotides in length; 2) hybridizes specifically to a nucleic acid having a sequence complementary to the DNA sequence set forth in SEQ ID NO:1.;
- f) removing any unbound labeled nucleic acid probe; and
- g) detecting the presence of labeled nucleic acid probe hybridized to the amplification product so as to thereby detect the presence of a nucleic acid encoding a prostate specific membrane antigen in a subject.

102. A method of detecting the presence of a nucleic acid encoding a prostate specific membrane antigen in a subject which comprises:

- a) obtaining a suitable sample of mRNA from the subject;
- b) generating a double-stranded cDNA from the mRNA;
- c) contacting the double-stranded cDNA from (b) with one primer having a sequence which is complementary to a portion of the sequence set forth in SEQ ID NO:1 and a

Applicants: Ron S. Israeli et al.  
Serial No.: 08/466,381  
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- second primer having a sequence which comprises a different portion of the sequence set forth in SEQ ID NO:1;
- d) amplifying the double stranded cDNA using a polymerase chain reaction to obtain an amplification product;
  - e) contacting the amplification product of (d) under hybridizing conditions with a labeled nucleic acid probe which 1) is at least 15 nucleotides in length; 2) hybridizes specifically to a nucleic acid having a sequence set forth in SEQ ID NO:1.;
  - f) removing any unbound labeled nucleic acid probe; and
  - g) detecting the presence of labeled nucleic acid probe hybridized to the amplification product so as to thereby detect the presence of a nucleic acid encoding a prostate specific membrane antigen in a subject.

103. The method of any one of claims 97-102, wherein the sample is blood, lymph nodes, or bone marrow.